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<b>(54) Title:</b> MODIFIED PEPTIDES AS THERAPEUTIC AGENTS			
<b>(57) Abstract</b> <p>The present invention concerns fusion of Fc domains with biologically active peptides and a process for preparing pharmaceutical agents using biologically active peptides. In this invention, pharmacologically active compounds are prepared by a process comprising: a) selecting at least one peptide that modulates the activity of a protein of interest; and b) preparing a pharmacologic agent comprising an Fc domain covalently linked to at least one amino acid of the selected peptide. Linkage to the vehicle increases the half-life of the peptide, which otherwise would be quickly degraded <i>in vivo</i>. The preferred vehicle is an Fc domain. The peptide is preferably selected by phage display, <i>E. coli</i> display, ribosome display, RNA-peptide screening, or chemical-peptide screening.</p>			

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## Modified Peptides as Therapeutic Agents

### Background of the Invention

Recombinant proteins are an emerging class of therapeutic agents.

5 Such recombinant therapeutics have engendered advances in protein formulation and chemical modification. Such modifications can protect therapeutic proteins, primarily by blocking their exposure to proteolytic enzymes. Protein modifications may also increase the therapeutic protein's stability, circulation time, and biological activity. A review  
10 article describing protein modification and fusion proteins is Francis (1992), Focus on Growth Factors 3:4-10 (Mediscript, London), which is hereby incorporated by reference.

One useful modification is combination with the "Fc" domain of an antibody. Antibodies comprise two functionally independent parts, a  
15 variable domain known as "Fab", which binds antigen, and a constant domain known as "Fc", which links to such effector functions as complement activation and attack by phagocytic cells. An Fc has a long serum half-life, whereas an Fab is short-lived. Capon et al. (1989), Nature 337: 525-31. When constructed together with a therapeutic protein, an Fc  
20 domain can provide longer half-life or incorporate such functions as Fc receptor binding, protein A binding, complement fixation and perhaps even placental transfer. Id. Table 1 summarizes use of Fc fusions known in the art.

Table 1—Fc fusion with therapeutic proteins

Form of Fc	Fusion partner	Therapeutic implications	Reference
IgG1	N-terminus of CD30-L	Hodgkin's disease; anaplastic lymphoma; T-cell leukemia	U.S. Patent No. 5,480,981
Murine Fcγ2a	IL-10	anti-inflammatory; transplant rejection	Zheng <i>et al.</i> (1995), <i>J. Immunol.</i> 154: 5590-600
IgG1	TNF receptor	septic shock	Fisher <i>et al.</i> (1996), <i>N. Engl. J. Med.</i> 334: 1697-1702; Van Zee, K. <i>et al.</i> (1996), <i>J. Immunol.</i> 156: 2221-30
IgG, IgA, IgM, or IgE (excluding the first domain)	TNF receptor	inflammation, autoimmune disorders	U.S. Pat. No. 5,808,029, issued September 15, 1998
IgG1	CD4 receptor	AIDS	Capon <i>et al.</i> (1989), <i>Nature</i> 337: 525-31
IgG1, IgG3	N-terminus of IL-2	anti-cancer, antiviral	Harvill <i>et al.</i> (1995), <i>Immunotech.</i> 1: 95-105
IgG1	C-terminus of OPG	osteoarthritis; bone density	WO 97/23614, published July 3, 1997
IgG1	N-terminus of leptin	anti-obesity	PCT/US 97/23183, filed December 11, 1997
Human Ig Cy1	CTLA-4	autoimmune disorders	Linsley (1991), <i>J. Exp. Med.</i> 174:561-9

A much different approach to development of therapeutic agents is peptide library screening. The interaction of a protein ligand with its receptor often takes place at a relatively large interface. However, as demonstrated for human growth hormone and its receptor, only a few key residues at the interface contribute to most of the binding energy. Clackson *et al.* (1995), *Science* 267: 383-6. The bulk of the protein ligand merely displays the binding epitopes in the right topology or serves functions unrelated to binding. Thus, molecules of only "peptide" length (2 to 40 amino acids) can bind to the receptor protein of a given large protein ligand. Such peptides may mimic the bioactivity of the large protein ligand ("peptide agonists") or, through competitive binding, inhibit the bioactivity of the large protein ligand ("peptide antagonists").

Phage display peptide libraries have emerged as a powerful method in identifying such peptide agonists and antagonists. See, for example, Scott *et al.* (1990), *Science* 249: 386; Devlin *et al.* (1990), *Science* 249: 404; U.S. Pat. No. 5,223,409, issued June 29, 1993; U.S. Pat. No. 5,733,731, issued March 31, 1998; U.S. Pat. No. 5,498,530, issued March 12, 1996; U.S. Pat. No. 5,432,018, issued July 11, 1995; U.S. Pat. No. 5,338,665, issued August 16, 1994; U.S. Pat. No. 5,922,545, issued July 13, 1999; WO 96/40987, published December 19, 1996; and WO 98/15833, published April 16, 1998 (each of which is incorporated by reference). In such libraries, random peptide sequences are displayed by fusion with coat proteins of filamentous phage. Typically, the displayed peptides are affinity-eluted against an antibody-immobilized extracellular domain of a receptor. The retained phages may be enriched by successive rounds of affinity purification and repropagation. The best binding peptides may be sequenced to identify key residues within one or more structurally related families of peptides. See, e.g., Cwirla *et al.* (1997), *Science* 276: 1696-9, in which two distinct families were identified. The peptide sequences may also suggest which residues may be safely replaced by alanine scanning or by mutagenesis at the DNA level. Mutagenesis libraries may be created and screened to further optimize the sequence of the best binders. Lowman (1997), *Ann. Rev. Biophys. Biomol. Struct.* 26: 401-24.

Structural analysis of protein-protein interaction may also be used to suggest peptides that mimic the binding activity of large protein ligands. In such an analysis, the crystal structure may suggest the identity and relative orientation of critical residues of the large protein ligand, from which a peptide may be designed. See, e.g., Takasaki *et al.* (1997), *Nature Biotech.* 15: 1266-70. These analytical methods may also be used to investigate the interaction between a receptor protein and peptides

selected by phage display, which may suggest further modification of the peptides to increase binding affinity.

Other methods compete with phage display in peptide research. A peptide library can be fused to the carboxyl terminus of the lac repressor and expressed in E. coli. Another E. coli-based method allows display on the cell's outer membrane by fusion with a peptidoglycan-associated lipoprotein (PAL). Hereinafter, these and related methods are collectively referred to as "E. coli display." In another method, translation of random RNA is halted prior to ribosome release, resulting in a library of polypeptides with their associated RNA still attached. Hereinafter, this and related methods are collectively referred to as "ribosome display." Other methods employ chemical linkage of peptides to RNA; see, for example, Roberts & Szostak (1997), Proc. Natl. Acad. Sci. USA, 94: 12297-303. Hereinafter, this and related methods are collectively referred to as "RNA-peptide screening." Chemically derived peptide libraries have been developed in which peptides are immobilized on stable, non-biological materials, such as polyethylene rods or solvent-permeable resins. Another chemically derived peptide library uses photolithography to scan peptides immobilized on glass slides. Hereinafter, these and related methods are collectively referred to as "chemical-peptide screening." Chemical-peptide screening may be advantageous in that it allows use of D-amino acids and other unnatural analogues, as well as non-peptide elements. Both biological and chemical methods are reviewed in Wells & Lowman (1992), Curr. Opin. Biotechnol. 3: 355-62.

Conceptually, one may discover peptide mimetics of any protein using phage display and the other methods mentioned above. These methods have been used for epitope mapping, for identification of critical amino acids in protein-protein interactions, and as leads for the discovery of new therapeutic agents. E.g., Cortese et al. (1996), Curr. Opin. Biotech. 7:

616-21. Peptide libraries are now being used most often in immunological studies, such as epitope mapping. Kreeger (1996), The Scientist 10(13): 19-20.

5       Of particular interest here is use of peptide libraries and other techniques in the discovery of pharmacologically active peptides. A number of such peptides identified in the art are summarized in Table 2. The peptides are described in the listed publications, each of which is hereby incorporated by reference. The pharmacologic activity of the peptides is described, and in many instances is followed by a shorthand  
10       term therefor in parentheses. Some of these peptides have been modified (e.g., to form C-terminally cross-linked dimers). Typically, peptide libraries were screened for binding to a receptor for a pharmacologically active protein (e.g., EPO receptor). In at least one instance (CTLA4), the peptide library was screened for binding to a monoclonal antibody.

Table 2—Pharmacologically active peptides

Form of peptide	Binding partner/ protein of interest <sup>a</sup>	Pharmacologic activity	Reference
intrapeptide disulfide-bonded	EPO receptor	EPO-mimetic	Wrighton <i>et al.</i> (1996), <i>Science</i> 273: 458-63; U.S. Pat. No. 5,773,569, issued June 30, 1998 to Wrighton <i>et al.</i>
C-terminally cross-linked dimer	EPO receptor	EPO-mimetic	Livnah <i>et al.</i> (1996), <i>Science</i> 273: 464-71; Wrighton <i>et al.</i> (1997), <i>Nature Biotechnology</i> 15: 1261-5; International patent application WO 96/40772, published Dec. 19, 1996
linear	EPO receptor	EPO-mimetic	Naranda <i>et al.</i> (1999), <i>Proc. Natl. Acad. Sci. USA</i> , 96: 7569-74
linear	c-Mpl	TPO-mimetic	Cwirla <i>et al.</i> (1997) <i>Science</i> 276: 1696-9; U.S. Pat. No. 5,869,451, issued Feb. 9, 1999; U.S. Pat. No. 5,932,946, issued Aug. 3, 1999
C-terminally cross-linked dimer	c-Mpl	TPO-mimetic	Cwirla <i>et al.</i> (1997), <i>Science</i> 276: 1696-9
disulfide-linked dimer		stimulation of hematopoiesis ("G-CSF-mimetic")	Paukovits <i>et al.</i> (1984), <i>Hoppe-Seyler's Z. Physiol. Chem.</i> 365: 303-11; Laerum <i>et al.</i> (1988), <i>Exp. Hemat.</i> 16: 274-80
alkylene-linked dimer		G-CSF-mimetic	Bhatnagar <i>et al.</i> (1996), <i>J. Med. Chem.</i> 39: 3814-9; Cuthbertson <i>et al.</i> (1997), <i>J. Med. Chem.</i> 40: 2876-82; King <i>et al.</i> (1991), <i>Exp. Hematol.</i> 19:481; King <i>et al.</i> (1995), <i>Blood</i> 86 (Suppl. 1): 309a
linear	IL-1 receptor	inflammatory and autoimmune diseases ("IL-1 antagonist" or "IL-1ra-mimetic")	U.S. Pat. No. 5,608,035; U.S. Pat. No. 5,786,331; U.S. Pat. No. 5,880,096; Yanofsky <i>et al.</i> (1996),

<sup>a</sup> The protein listed in this column may be bound by the associated peptide (e.g., EPO receptor, IL-1 receptor) or mimicked by the associated peptide. The references listed for each clarify whether the molecule is bound by or mimicked by the peptides.



			Proc. Natl. Acad. Sci. 93: 7381-6; Akeson <i>et al.</i> (1996), <i>J. Biol. Chem.</i> 271: 30517-23; Wieczorek <i>et al.</i> (1997), <i>Pol. J. Pharmacol.</i> 49: 107-17; Yanofsky (1996), <i>PNAs</i> , 93:7381-7386.
linear	Facteur thymique serique (FTS)	stimulation of lymphocytes ("FTS-mimetic")	Inagaki-Ohara <i>et al.</i> (1996), <i>Cellular Immunol.</i> 171: 30-40; Yoshida (1984), <i>Int. J. Immunopharmacol.</i> 6:141-6.
intrapeptide disulfide bonded	CTLA4 MAb	CTLA4-mimetic	Fukumoto <i>et al.</i> (1998), <i>Nature Biotech.</i> 16: 267-70
exocyclic	TNF- $\alpha$ receptor	TNF- $\alpha$ antagonist	Takasaki <i>et al.</i> (1997), <i>Nature Biotech.</i> 15:1266-70; WO 98/53842, published December 3, 1998
linear	TNF- $\alpha$ receptor	TNF- $\alpha$ antagonist	Chirinos-Rojas ( ), <i>J. Imm.</i> , 5621-5626.
intrapeptide disulfide bonded	C3b	inhibition of complement activation; autoimmune diseases ("C3b-antagonist")	Sahu <i>et al.</i> (1996), <i>J. Immunol.</i> 157: 884-91; Morikis <i>et al.</i> (1998), <i>Protein Sci.</i> 7: 619-27
linear	vinculin	cell adhesion processes—cell growth, differentiation, wound healing, tumor metastasis ("vinculin binding")	Adey <i>et al.</i> (1997), <i>Biochem. J.</i> 324: 523-8
linear	C4 binding protein (C4BP)	anti-thrombotic	Linse <i>et al.</i> (1997), <i>J. Biol. Chem.</i> 272: 14658-65
linear	urokinase receptor	processes associated with urokinase interaction with its receptor (e.g., angiogenesis, tumor cell invasion and metastasis); ("UKR antagonist")	Goodson <i>et al.</i> (1994), <i>Proc. Natl. Acad. Sci.</i> 91: 7129-33; International application WO 97/35969, published October 2, 1997
linear	Mdm2, Hdm2	Inhibition of inactivation of p53 mediated by Mdm2 or hdm2; anti-tumor ("Mdm/hdm antagonist")	Picksley <i>et al.</i> (1994), <i>Oncogene</i> 9: 2523-9; Bottger <i>et al.</i> (1997) <i>J. Mol. Biol.</i> 269: 744-56; Bottger <i>et al.</i> (1996), <i>Oncogene</i> 13: 2141-7
linear	p21 <sup>WAF1</sup>	anti-tumor by mimicking the activity of p21 <sup>WAF1</sup>	Ball <i>et al.</i> (1997), <i>Curr. Biol.</i> 7: 71-80
linear	farnesyl	anti-cancer by preventing	Gibbs <i>et al.</i> (1994), <i>Cell</i>

<sup>b</sup> FTS is a thymic hormone mimicked by the molecule of this invention rather than a receptor bound by the molecule of this invention.

linear	transferase Ras effector domain	activation of ras oncogene anti-cancer by inhibiting biological function of the ras oncogene	77:175-178 Moodie et al. (1994), <u>Trends Genet</u> 10: 44-48 Rodriguez et al. (1994), <u>Nature</u> 370:527-532
linear	SH2/SH3 domains	anti-cancer by inhibiting tumor growth with activated tyrosine kinases	Pawson et al (1993), <u>Curr. Biol.</u> 3:434-432 Yu et al. (1994), <u>Cell</u> 76:933-945
linear	p16 <sup>INK4</sup>	anti-cancer by mimicking activity of p16; e.g., inhibiting cyclin D-Cdk complex ("p16-mimetic")	Fähraeus et al. (1996), <u>Curr. Biol.</u> 6:84-91
linear	Src, Lyn	inhibition of Mast cell activation, IgE-related conditions, type I hypersensitivity ("Mast cell antagonist")	Stauffer et al. (1997), <u>Biochem.</u> 36: 9388-94
linear	Mast cell protease	treatment of inflammatory disorders mediated by release of tryptase-6 ("Mast cell protease inhibitors")	International application WO 98/33812, published August 6, 1998
linear	SH3 domains	treatment of SH3- mediated disease states ("SH3 antagonist")	Rickles et al. (1994), <u>EMBO J.</u> 13: 5598-5604; Sparks et al. (1994), <u>J.</u> <u>Biol. Chem.</u> 269: 23853- 6; Sparks et al. (1996), <u>Proc. Natl. Acad. Sci.</u> 93: 1540-4
linear	HBV core antigen (HBcAg)	treatment of HBV viral infections ("anti-HBV")	Dyson & Muray (1995), <u>Proc. Natl. Acad. Sci.</u> 92: 2194-8
linear	selectins	neutrophil adhesion; inflammatory diseases ("selectin antagonist")	Martens et al. (1995), <u>J.</u> <u>Biol. Chem.</u> 270: 21129- 36; European patent application EP 0 714 912, published June 5, 1996
linear, cyclized	calmodulin	calmodulin antagonist	Pierce et al. (1995), <u>Molec. Diversity</u> 1: 259- 65; Dedman et al. (1993), <u>J. Biol. Chem.</u> 268: 23025-30; Adey & Kay (1996), <u>Gene</u> 169: 133-4
linear, cyclized-	integrins	tumor-homing; treatment for conditions related to integrin-mediated cellular events, including platelet aggregation, thrombosis, wound healing, osteoporosis, tissue repair, angiogenesis (e.g.,	International applications WO 95/14714, published June 1, 1995; WO 97/08203, published March 6, 1997; WO 98/10795, published March 19, 1998; WO 99/24462, published May

		for treatment of cancer), and tumor invasion ("integrin-binding")	20, 1999; Kraft <i>et al.</i> (1999), <i>J. Biol. Chem.</i> 274: 1979-1985
cyclic, linear	fibronectin and extracellular matrix components of T cells and macrophages	treatment of inflammatory and autoimmune conditions	WO 98/09985, published March 12, 1998
linear	somatostatin and cortistatin	treatment or prevention of hormone-producing tumors, acromegaly, giantism, dementia, gastric ulcer, tumor growth, inhibition of hormone secretion, modulation of sleep or neural activity	European patent application 0 911 393, published April 28, 1999
linear	bacterial lipopolysac- charide	antibiotic; septic shock; disorders modulatable by CAP37	U.S. Pat. No. 5,877,151, issued March 2, 1999
linear or cyclic, including D- amino acids	pardaxin, mellitin	antipathogenic	WO 97/31019, published 28 August 1997
linear, cyclic	VIP	impotence, neurodegenerative disorders	WO 97/40070, published October 30, 1997
linear	CTLs	cancer	EP 0 770 624, published May 2, 1997
linear	THF-gamma2		Burnstein (1988), <i>Biochem.</i> , 27:4066-71.
linear	Amylin		Cooper (1987), <i>Proc.</i> <i>Natl. Acad. Sci.</i> , 84:8628-32.
linear	Adrenomedullin		Kitamura (1993), <i>BBRC</i> , 192:553-60.
cyclic, linear	VEGF	anti-angiogenic; cancer, rheumatoid arthritis, diabetic retinopathy, psoriasis ("VEGF antagonist")	Fairbrother (1998), <i>Biochem.</i> , 37:17754- 17764.
cyclic	MMP	inflammation and autoimmune disorders; tumor growth ("MMP inhibitor")	Koivunen (1999), <i>Nature</i> <i>Biotech.</i> , 17:768-774.
	HGH fragment		U.S. Pat. No. 5,869,452
	Echistatin	inhibition of platelet aggregation	Gan (1988), <i>J. Biol.</i> <i>Chem.</i> , 263:19827-32.
linear	SLE autoantibody	SLE	WO 96/30057, published October 3, 1996
	GD1alpha	suppression of tumor metastasis	Ishikawa <i>et al.</i> (1998), <i>FEBS Lett.</i> 441 (1): 20-4
	antiphospholipid	endothelial cell activation	Blank <i>et al.</i> (1999), <i>Proc.</i>

	beta-2-glycoprotein-I ( $\beta$ 2GPI) antibodies	antiphospholipid syndrome (APS), thromboembolic phenomena, thrombocytopenia, and recurrent fetal loss	Natl. Acad. Sci. USA 96: 5164-8
linear	T Cell Receptor beta chain	diabetes	WO 96/11214, published April 18, 1996

Peptides identified by peptide library screening have been regarded as "leads" in development of therapeutic agents rather than as therapeutic agents themselves. Like other proteins and peptides, they would be rapidly removed in vivo either by renal filtration, cellular clearance mechanisms in the reticuloendothelial system, or proteolytic degradation. Francis (1992), Focus on Growth Factors 3: 4-11. As a result, the art presently uses the identified peptides to validate drug targets or as scaffolds for design of organic compounds that might not have been as easily or as quickly identified through chemical library screening. Lowman (1997), Ann. Rev. Biophys. Biomol. Struct. 26: 401-24; Kay *et al.* (1998), Drug Disc. Today 3: 370-8. The art would benefit from a process by which such peptides could more readily yield therapeutic agents.

### Summary of the Invention

The present invention concerns a process by which the in vivo half-life of one or more biologically active peptides is increased by fusion with a vehicle. In this invention, pharmacologically active compounds are prepared by a process comprising:

- a) selecting at least one peptide that modulates the activity of a protein of interest; and
- b) preparing a pharmacologic agent comprising at least one vehicle covalently linked to at least one amino acid sequence of the selected peptide.

The preferred vehicle is an Fc domain. The peptides screened in step (a) are preferably expressed in a phage display library. The vehicle and the

peptide may be linked through the N- or C-terminus of the peptide or the vehicle, as described further below. Derivatives of the above compounds (described below) are also encompassed by this invention.

5 The compounds of this invention may be prepared by standard synthetic methods, recombinant DNA techniques, or any other methods of preparing peptides and fusion proteins. Compounds of this invention that encompass non-peptide portions may be synthesized by standard organic chemistry reactions, in addition to standard peptide chemistry reactions when applicable.

10 The primary use contemplated is as therapeutic or prophylactic agents. The vehicle-linked peptide may have activity comparable to—or even greater than—the natural ligand mimicked by the peptide. In addition, certain natural ligand-based therapeutic agents might induce antibodies against the patient's own endogenous ligand; the vehicle-linked  
15 peptide avoids this pitfall by having little or typically no sequence identity with the natural ligand.

Although mostly contemplated as therapeutic agents, compounds of this invention may also be useful in screening for such agents. For example, one could use an Fc-peptide (e.g., Fc-SH2 domain peptide) in an  
20 assay employing anti-Fc coated plates. The vehicle, especially Fc, may make insoluble peptides soluble and thus useful in a number of assays.

The compounds of this invention may be used for therapeutic or prophylactic purposes by formulating them with appropriate pharmaceutical carrier materials and administering an effective amount to  
25 a patient, such as a human (or other mammal) in need thereof. Other related aspects are also included in the instant invention.

Numerous additional aspects and advantages of the present invention will become apparent upon consideration of the figures and detailed description of the invention.

### Brief Description of the Figures

Figure 1 shows a schematic representation of an exemplary process of the invention. In this preferred process, the vehicle is an Fc domain, which is linked to the peptide covalently by expression from a DNA construct encoding both the Fc domain and the peptide. As noted in  
5 Figure 1, the Fc domains spontaneously form a dimer in this process.

Figure 2 shows exemplary Fc dimers that may be derived from an IgG1 antibody. "Fc" in the figure represents any of the Fc variants within the meaning of "Fc domain" herein. "X<sup>1</sup>" and "X<sup>2</sup>" represent peptides or  
10 linker-peptide combinations as defined hereinafter. The specific dimers are as follows:

A, D: Single disulfide-bonded dimers. IgG1 antibodies typically have two disulfide bonds at the hinge region between the constant and variable domains. The Fc domain in Figures 2A and 2D may be formed by  
15 truncation between the two disulfide bond sites or by substitution of a cysteinyl residue with an unreactive residue (e.g., alanyl). In Figure 2A, the Fc domain is linked at the amino terminus of the peptides; in 2D, at the carboxyl terminus.

B, E: Doubly disulfide-bonded dimers. This Fc domain may be  
20 formed by truncation of the parent antibody to retain both cysteinyl residues in the Fc domain chains or by expression from a construct including a sequence encoding such an Fc domain. In Figure 2B, the Fc domain is linked at the amino terminus of the peptides; in 2E, at the carboxyl terminus.

25 C, F: Noncovalent dimers. This Fc domain may be formed by elimination of the cysteinyl residues by either truncation or substitution. One may desire to eliminate the cysteinyl residues to avoid impurities formed by reaction of the cysteinyl residue with cysteinyl residues of other

proteins present in the host cell. The noncovalent bonding of the Fc domains is sufficient to hold together the dimer.

Other dimers may be formed by using Fc domains derived from different types of antibodies (e.g., IgG2, IgM).

5           Figure 3 shows the structure of preferred compounds of the invention that feature tandem repeats of the pharmacologically active peptide. Figure 3A shows a single chain molecule and may also represent the DNA construct for the molecule. Figure 3B shows a dimer in which the linker-peptide portion is present on only one chain of the dimer. Figure 3C  
10 shows a dimer having the peptide portion on both chains. The dimer of Figure 3C will form spontaneously in certain host cells upon expression of a DNA construct encoding the single chain shown in Figure 3A. In other host cells, the cells could be placed in conditions favoring formation of dimers or the dimers can be formed in vitro.

15           Figure 4 shows exemplary nucleic acid and amino acid sequences (SEQ ID NOS: 1 and 2, respectively) of human IgG1 Fc that may be used in this invention.

Figure 5 shows a synthetic scheme for the preparation of PEGylated peptide 19 (SEQ ID NO: 3).

20           Figure 6 shows a synthetic scheme for the preparation of PEGylated peptide 20 (SEQ ID NO: 4).

Figure 7 shows the nucleotide and amino acid sequences (SEQ ID NOS: 5 and 6, respectively) of the molecule identified as "Fc-TMP" in Example 2 hereinafter.

25           Figure 8 shows the nucleotide and amino acid sequences (SEQ. ID. NOS: 7 and 8, respectively) of the molecule identified as "Fc-TMP-TMP" in Example 2 hereinafter.

Figure 9 shows the nucleotide and amino acid sequences (SEQ. ID. NOS: 9 and 10, respectively) of the molecule identified as "TMP-TMP-Fc" in Example 2 hereinafter.

Figure 10 shows the nucleotide and amino acid sequences (SEQ. ID. NOS: 11 and 12, respectively) of the molecule identified as "TMP-Fc" in Example 2 hereinafter.

Figure 11 shows the number of platelets generated in vivo in normal female BDF1 mice treated with one 100 µg/kg bolus injection of various compounds, with the terms defined as follows.

10 PEG-MGDF: 20 kD average molecular weight PEG attached by reductive amination to the N-terminal amino group of amino acids 1-163 of native human TPO, which is expressed in E. coli (so that it is not glycosylated);

15 TMP: the TPO-mimetic peptide having the amino acid sequence IEGPTLRQWLAARA (SEQ ID NO: 13);

TMP-TMP: the TPO-mimetic peptide having the amino acid sequence IEGPTLRQWLAARA-GGGGGGGG-IEGPTLRQWLAARA (SEQ ID NO: 14);

20 PEG-TMP-TMP: the peptide of SEQ ID NO: 14, wherein the PEG group is a 5 kD average molecular weight PEG attached as shown in Figure 6;

Fc-TMP-TMP: the compound of SEQ ID NO: 8 (Figure 8) dimerized with an identical second monomer (i.e., Cys residues 7 and 10 are bound to the corresponding Cys residues in the second monomer to form a dimer, as shown in Figure 2); and

25 TMP-TMP-Fc is the compound of SEQ ID NO: 10 (Figure 9) dimerized in the same way as TMP-TMP-Fc except that the Fc domain is attached at the C-terminal end rather than the N-terminal end of the TMP-TMP peptide.



Figure 12 shows the number of platelets generated in vivo in normal BDF1 mice treated with various compounds delivered via implanted osmotic pumps over a 7-day period. The compounds are as defined for Figure 7.

5        Figure 13 shows the nucleotide and amino acid sequences (SEQ. ID. NOS: 15 and 16, respectively) of the molecule identified as "Fc-EMP" in Example 3 hereinafter.

Figure 14 shows the nucleotide and amino acid sequences (SEQ ID NOS: 17 and 18, respectively) of the molecule identified as "EMP-Fc" in  
10    Example 3 hereinafter.

Figure 15 shows the nucleotide and amino acid sequences (SEQ ID NOS: 19 and 20, respectively) of the molecule identified as "EMP-EMP-Fc" in Example 3 hereinafter.

Figure 16 shows the nucleotide and amino acid sequences (SEQ ID NOS: 21 and 22, respectively) of the molecule identified as "Fc-EMP-EMP"  
15    in Example 3 hereinafter.

Figures 17A and 17B show the DNA sequence (SEQ ID NO: 23) inserted into pCFM1656 between the unique AatII (position #4364 in pCFM1656) and SacII (position #4585 in pCFM1656) restriction sites to  
20    form expression plasmid pAMG21 (ATCC accession no. 98113).

Figure 18A shows the hemoglobin, red blood cells, and hematocrit generated in vivo in normal female BDF1 mice treated with one 100 µg/kg bolus injection of various compounds. Figure 18B shows the same results with mice treated with 100 µg/kg per day delivered ~~the same dose~~ by 7-  
25    day micro-osmotic pump with the EMPs delivered at 100 µg/kg, rhEPO at 30U/mouse. (In both experiments, neutrophils, lymphocytes, and platelets were unaffected.) In these figures, the terms are defined as follows.

Fc-EMP: the compound of SEQ ID NO: 16 (Figure 13) dimerized with an identical second monomer (i.e., Cys residues 7 and 10 are

bound to the corresponding Cys residues in the second monomer to form a dimer, as shown in Figure 2);

EMP-Fc: the compound of SEQ ID NO: 18 (Figure 14) dimerized in the same way as Fc-EMP except that the Fc domain is attached at the C-terminal end rather than the N-terminal end of the EMP peptide.

"EMP-EMP-Fc" refers to a tandem repeat of the same peptide (SEQ ID NO: 20) attached to the same Fc domain by the carboxyl terminus of the peptides. "Fc-EMP-EMP" refers to the same tandem repeat of the peptide but with the same Fc domain attached at the amino terminus of the tandem repeat. All molecules are expressed in E. coli and so are not glycosylated.

Figures 19A and 19B show the nucleotide and amino acid sequences (SEQ ID NOS: 1055 and 1056) of the Fc-TNF- $\alpha$  inhibitor fusion molecule described in Example 4 hereinafter.

Figures 20A and 20B show the nucleotide and amino acid sequences (SEQ ID NOS: 1057 and 1058) of the TNF- $\alpha$  inhibitor-Fc fusion molecule described in Example 4 hereinafter.

Figures 21A and 21B show the nucleotide and amino acid sequences (SEQ ID NOS: 1059 and 1060) of the Fc-IL-1 antagonist fusion molecule described in Example 5 hereinafter.

Figures 22A and 22B show the nucleotide and amino acid sequences (SEQ ID NOS: 1061 and 1062) of the IL-1 antagonist-Fc fusion molecule described in Example 5 hereinafter.

Figures 23A, 23B, and 23C show the nucleotide and amino acid sequences (SEQ ID NOS: 1063 and 1064) of the Fc-VEGF antagonist fusion molecule described in Example 6 hereinafter.

Figures 24A and 24B show the nucleotide and amino acid sequences (SEQ ID NOS: 1065 and 1066) of the VEGF antagonist-Fc fusion molecule described in Example 6 hereinafter.

Figures 25A and 25B show the nucleotide and amino acid sequences (SEQ ID NOS: 1067 and 1068) of the Fc-MMP inhibitor fusion molecule described in Example 7 hereinafter.

Figures 26A and 26B show the nucleotide and amino acid sequences (SEQ ID NOS: 1069 and 1070) of the MMP inhibitor-Fc fusion molecule described in Example 7 hereinafter.

## **Detailed Description of the Invention**

### **Definition of Terms**

The terms used throughout this specification are defined as follows, unless otherwise limited in specific instances.

The term "comprising" means that a compound may include additional amino acids on either or both of the N- or C- termini of the given sequence. Of course, these additional amino acids should not significantly interfere with the activity of the compound.

The term "vehicle" refers to a molecule that prevents degradation and/or increases half-life, reduces toxicity, reduces immunogenicity, or increases biological activity of a therapeutic protein. Exemplary vehicles include an Fc domain (which is preferred) as well as a linear polymer (e.g., polyethylene glycol (PEG), polylysine, dextran, etc.); a branched-chain polymer (see, for example, U.S. Patent No. 4,289,872 to Denkenwalter et al., issued September 15, 1981; 5,229,490 to Tam, issued July 20, 1993; WO 93/21259 by Frechet et al., published 28 October 1993); a lipid; a cholesterol group (such as a steroid); a carbohydrate or oligosaccharide; or any natural or synthetic protein, polypeptide or peptide that binds to a salvage receptor. Vehicles are further described hereinafter.

The term "native Fc" refers to molecule or sequence comprising the sequence of a non-antigen-binding fragment resulting from digestion of whole antibody, whether in monomeric or multimeric form. The original immunoglobulin source of the native Fc is preferably of human origin and  
5 may be any of the immunoglobulins, although IgG1 and IgG2 are preferred. Native Fc's are made up of monomeric polypeptides that may be linked into dimeric or multimeric forms by covalent (i.e., disulfide bonds) and non-covalent association. The number of intermolecular disulfide bonds between monomeric subunits of native Fc molecules  
10 ranges from 1 to 4 depending on class (e.g., IgG, IgA, IgE) or subclass (e.g., IgG1, IgG2, IgG3, IgA1, IgGA2). One example of a native Fc is a disulfide-bonded dimer resulting from papain digestion of an IgG (see Ellison *et al.* (1982), Nucleic Acids Res. 10: 4071-9). The term "native Fc" as used herein is generic to the monomeric, dimeric, and multimeric forms.

15 The term "Fc variant" refers to a molecule or sequence that is modified from a native Fc but still comprises a binding site for the salvage receptor, FcRn. International applications WO 97/34631 (published 25 September 1997) and WO 96/32478 describe exemplary Fc variants, as well as interaction with the salvage receptor, and are hereby incorporated  
20 by reference. Thus, the term "Fc variant" comprises a molecule or sequence that is humanized from a non-human native Fc. Furthermore, a native Fc comprises sites that may be removed because they provide structural features or biological activity that are not required for the fusion molecules of the present invention. Thus, the term "Fc variant" comprises  
25 a molecule or sequence that lacks one or more native Fc sites or residues that affect or are involved in (1) disulfide bond formation, (2) incompatibility with a selected host cell (3) N-terminal heterogeneity upon expression in a selected host cell, (4) glycosylation, (5) interaction with complement, (6) binding to an Fc receptor other than a salvage receptor, or

(7) antibody-dependent cellular cytotoxicity (ADCC). Fc variants are described in further detail hereinafter.

The term "Fc domain" encompasses native Fc and Fc variant molecules and sequences as defined above. As with Fc variants and native Fc's, the term "Fc domain" includes molecules in monomeric or multimeric form, whether digested from whole antibody or produced by other means.

The term "multimer" as applied to Fc domains or molecules comprising Fc domains refers to molecules having two or more polypeptide chains associated covalently, noncovalently, or by both covalent and non-covalent interactions. IgG molecules typically form dimers; IgM, pentamers; IgD, dimers; and IgA, monomers, dimers, trimers, or tetramers. Multimers may be formed by exploiting the sequence and resulting activity of the native Ig source of the Fc or by derivatizing (as defined below) such a native Fc.

The term "dimer" as applied to Fc domains or molecules comprising Fc domains refers to molecules having two polypeptide chains associated covalently or non-covalently. Thus, exemplary dimers within the scope of this invention are as shown in Figure 2.

The terms "derivatizing" and "derivative" or "derivatized" comprise processes and resulting compounds respectively in which (1) the compound has a cyclic portion; for example, cross-linking between cysteinyl residues within the compound; (2) the compound is cross-linked or has a cross-linking site; for example, the compound has a cysteinyl residue and thus forms cross-linked dimers in culture or in vivo; (3) one or more peptidyl linkage is replaced by a non-peptidyl linkage; (4) the N-terminus is replaced by  $-NRR^1$ ,  $NRC(O)R^1$ ,  $-NRC(O)OR^1$ ,  $-NRS(O)_2R^1$ ,  $-NHC(O)NHR$ , a succinimide group, or substituted or unsubstituted benzyloxycarbonyl-NH-, wherein R and  $R^1$  and the ring substituents are

as defined hereinafter; (5) the C-terminus is replaced by  $-C(O)R^2$  or  $-NR^3R^4$  wherein  $R^2$ ,  $R^3$  and  $R^4$  are as defined hereinafter; and (6) compounds in which individual amino acid moieties are modified through treatment with agents capable of reacting with selected side chains or terminal  
5 residues. Derivatives are further described hereinafter.

The term "peptide" refers to molecules of 2 to 40 amino acids, with molecules of 3 to 20 amino acids preferred and those of 6 to 15 amino acids most preferred. Exemplary peptides may be randomly generated by any of the methods cited above, carried in a peptide library (e.g., a phage  
10 display library), or derived by digestion of proteins.

The term "randomized" as used to refer to peptide sequences refers to fully random sequences (e.g., selected by phage display methods) and sequences in which one or more residues of a naturally occurring molecule is replaced by an amino acid residue not appearing in that position in the  
15 naturally occurring molecule. Exemplary methods for identifying peptide sequences include phage display, *E. coli* display, ribosome display, RNA-peptide screening, chemical screening, and the like.

The term "pharmacologically active" means that a substance so described is determined to have activity that affects a medical parameter  
20 (e.g., blood pressure, blood cell count, cholesterol level) or disease state (e.g., cancer, autoimmune disorders). Thus, pharmacologically active peptides comprise agonistic or mimetic and antagonistic peptides as defined below.

The terms "-mimetic peptide" and "-agonist peptide" refer to a  
25 peptide having biological activity comparable to a protein (e.g., EPO, TPO, G-CSF) that interacts with a protein of interest. These terms further include peptides that indirectly mimic the activity of a protein of interest, such as by potentiating the effects of the natural ligand of the protein of interest; see, for example, the G-CSF-mimetic peptides listed in Tables 2

and 7. Thus, the term "EPO-mimetic peptide" comprises any peptides that can be identified or derived as described in Wrighton et al. (1996), Science 273: 458-63, Naranda et al. (1999), Proc. Natl. Acad. Sci. USA 96: 7569-74, or any other reference in Table 2 identified as having EPO-mimetic subject matter. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually disclosed therein by following the disclosed procedures with different peptide libraries.

The term "TPO-mimetic peptide" comprises peptides that can be identified or derived as described in Cwirla et al. (1997), Science 276: 1696-9, U.S. Pat. Nos. 5,869,451 and 5,932,946 and any other reference in Table 2 identified as having TPO-mimetic subject matter, as well as the U.S. patent application, "Thrombopoietic Compounds," filed on even date herewith and hereby incorporated by reference. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually disclosed therein by following the disclosed procedures with different peptide libraries.

The term "G-CSF-mimetic peptide" comprises any peptides that can be identified or described in Paukovits et al. (1984), Hoppe-Seylers Z. Physiol. Chem. 365: 303-11 or any of the references in Table 2 identified as having G-CSF-mimetic subject matter. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually disclosed therein by following the disclosed procedures with different peptide libraries.

The term "CTLA4-mimetic peptide" comprises any peptides that can be identified or derived as described in Fukumoto et al. (1998), Nature Biotech. 16: 267-70. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually

disclosed therein by following the disclosed procedures with different peptide libraries.

The term "-antagonist peptide" or "inhibitor peptide" refers to a peptide that blocks or in some way interferes with the biological activity of the associated protein of interest, or has biological activity comparable to a known antagonist or inhibitor of the associated protein of interest. Thus, the term "TNF-antagonist peptide" comprises peptides that can be identified or derived as described in Takasaki *et al.* (1997), *Nature Biotech.* 15: 1266-70 or any of the references in Table 2 identified as having TNF-antagonistic subject matter. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually disclosed therein by following the disclosed procedures with different peptide libraries.

The terms "IL-1 antagonist" and "IL-1ra-mimetic peptide" comprises peptides that inhibit or down-regulate activation of the IL-1 receptor by IL-1. IL-1 receptor activation results from formation of a complex among IL-1, IL-1 receptor, and IL-1 receptor accessory protein. IL-1 antagonist or IL-1ra-mimetic peptides bind to IL-1, IL-1 receptor, or IL-1 receptor accessory protein and obstruct complex formation among any two or three components of the complex. Exemplary IL-1 antagonist or IL-1ra-mimetic peptides can be identified or derived as described in U.S. Pat. Nos. 5,608,035, 5,786,331, 5,880,096, or any of the references in Table 2 identified as having IL-1ra-mimetic or IL-1 antagonistic subject matter. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually disclosed therein by following the disclosed procedures with different peptide libraries.

The term "VEGF-antagonist peptide" comprises peptides that can be identified or derived as described in Fairbrother (1998), *Biochem.* 37:



17754-64, and in any of the references in Table 2 identified as having VEGF-antagonistic subject matter. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually disclosed therein by following the disclosed procedures with different peptide libraries.

The term "MMP inhibitor peptide" comprises peptides that can be identified or derived as described in Koivunen (1999), Nature Biotech. 17: 768-74 and in any of the references in Table 2 identified as having MMP inhibitory subject matter. Those of ordinary skill in the art appreciate that each of these references enables one to select different peptides than actually disclosed therein by following the disclosed procedures with different peptide libraries.

Additionally, physiologically acceptable salts of the compounds of this invention are also encompassed herein. By "physiologically acceptable salts" is meant any salts that are known or later discovered to be pharmaceutically acceptable. Some specific examples are: acetate; trifluoroacetate; hydrohalides, such as hydrochloride and hydrobromide; sulfate; citrate; tartrate; glycolate; and oxalate.

### Structure of compounds

In General. In the compositions of matter prepared in accordance with this invention, the peptide may be attached to the vehicle through the peptide's N-terminus or C-terminus. Thus, the vehicle-peptide molecules of this invention may be described by the following formula I:

I



wherein:

F<sup>1</sup> is a vehicle (preferably an Fc domain);

X<sup>1</sup> and X<sup>2</sup> are each independently selected from -(L<sup>1</sup>)<sub>c</sub>-P<sup>1</sup>, -(L<sup>1</sup>)<sub>c</sub>-P<sup>1</sup>-(L<sup>2</sup>)<sub>d</sub>-P<sup>2</sup>, -(L<sup>1</sup>)<sub>c</sub>-P<sup>1</sup>-(L<sup>2</sup>)<sub>d</sub>-P<sup>2</sup>-(L<sup>3</sup>)<sub>e</sub>-P<sup>3</sup>, and -(L<sup>1</sup>)<sub>c</sub>-P<sup>1</sup>-(L<sup>2</sup>)<sub>d</sub>-P<sup>2</sup>-(L<sup>3</sup>)<sub>e</sub>-P<sup>3</sup>-(L<sup>4</sup>)<sub>f</sub>-P<sup>4</sup>

}}}

$P^1$ ,  $P^2$ ,  $P^3$ , and  $P^4$  are each independently sequences of pharmacologically active peptides;

$L^1$ ,  $L^2$ ,  $L^3$ , and  $L^4$  are each independently linkers; and

a, b, c, d, e, and f are each independently 0 or 1, provided that at least one of a and b is 1.

Thus, compound I comprises preferred compounds of the formulae

II



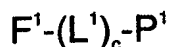
and multimers thereof wherein  $F^1$  is an Fc domain and is attached at the C-terminus of  $X^1$ ;

III



and multimers thereof wherein  $F^1$  is an Fc domain and is attached at the N-terminus of  $X^2$ ;

IV



and multimers thereof wherein  $F^1$  is an Fc domain and is attached at the N-terminus of  $-(L^1)_c-P^1$ ; and

V



and multimers thereof wherein  $F^1$  is an Fc domain and is attached at the N-terminus of  $-L^1-P^1-L^2-P^2$ .

Peptides. Any number of peptides may be used in conjunction with the present invention. Of particular interest are peptides that mimic the activity of EPO, TPO, growth hormone, G-CSF, GM-CSF, IL-1ra, leptin, CTLA4, TRAIL, TGF- $\alpha$ , and TGF- $\beta$ . Peptide antagonists are also of interest, particularly those antagonistic to the activity of TNF, leptin, any of the interleukins (IL-1, 2, 3, ...), and proteins involved in complement activation (e.g., C3b). Targeting peptides are also of interest, including

tumor-homing peptides, membrane-transporting peptides, and the like. All of these classes of peptides may be discovered by methods described in the references cited in this specification and other references.

5 Phage display, in particular, is useful in generating peptides for use in the present invention. It has been stated that affinity selection from libraries of random peptides can be used to identify peptide ligands for any site of any gene product. Dedman *et al.* (1993), J. Biol. Chem. 268: 23025-30. Phage display is particularly well suited for identifying peptides that bind to such proteins of interest as cell surface receptors or any  
10 proteins having linear epitopes. Wilson *et al.* (1998), Can. J. Microbiol. 44: 313-29; Kay *et al.* (1998), Drug Disc. Today 3: 370-8. Such proteins are extensively reviewed in Herz *et al.* (1997), J. Receptor & Signal Transduction Res. 17(5): 671-776, which is hereby incorporated by reference. Such proteins of interest are preferred for use in this invention.

15 A particularly preferred group of peptides are those that bind to cytokine receptors. Cytokines have recently been classified according to their receptor code. See Inglot (1997), Archivum Immunologiae et Therapiae Experimentalis 45: 353-7, which is hereby incorporated by reference. Among these receptors, most preferred are the CKRs (family I in  
20 Table 3). The receptor classification appears in Table 3.

**Table 3—Cytokine Receptors Classified by Receptor Code**

Cytokines (ligands)		Receptor Type	
family	subfamily	family	subfamily
I. Hematopoietic cytokines	1. IL-2, IL-4, IL-7, IL-9, IL-13, IL-15 2. IL-3, IL-5, GM-CSF 3. IL-6, IL-11, IL-12, LIF, OSM, CNTF, leptin (OB) 4. G-CSF, EPO, TPO, PRL, GH 5. IL-17, HVS-IL-17	I. Cytokine R (CKR)	1. shared $\gamma$ Cr 2. shared GP 140 $\beta$ R 3. 3.shared RP 130 4. "single chain" R 5. other R <sup>c</sup>
II. IL-10 ligands	IL-10, BCRF-1, HSV-IL-10	II. IL-10 R	
III. Interferons	1. IFN- $\alpha$ , $\alpha$ 2, $\alpha$ 4, m, t, IFN- $\beta$ <sup>d</sup> 2. IFN- $\gamma$	III. Interferon R	1. IFNAR 2. IFNGR
IV. IL-1 ligands	1. IL-1 $\alpha$ , IL-1 $\beta$ , IL-1Ra	IV. IL-1R	
V. TNF ligands	1. TNF- $\alpha$ , TNF- $\beta$ (LT), FAS1, CD40 L, CD30L, CD27 L	V. NGF/TNF R <sup>e</sup>	
VI. Chemokines	1. $\alpha$ chemokines: IL-8, GRO $\alpha$ , $\beta$ , $\gamma$ , IF-10, PF-4, SDF-1 2. $\beta$ chemokines: MIP1 $\alpha$ , MIP1 $\beta$ , MCP-1,2,3,4, RANTES, eotaxin 3. $\gamma$ chemokines: lymphotactin	VI. Chemokine R	1. CXCR 2. CCR 3. CR 4. DARC <sup>f</sup>

<sup>c</sup> IL-17R belongs to the CKR family but is not assigned to any of the 4 indicated subfamilies.

<sup>d</sup> Other IFN type I subtypes remain unassigned. Hematopoietic cytokines, IL-10 ligands and interferons do not possess functional intrinsic protein kinases. The signaling molecules for the cytokines are JAK's, STATs and related non-receptor molecules. IL-14, IL-16 and IL-18 have been cloned but according to the receptor code they remain unassigned.

<sup>e</sup> TNF receptors use multiple, distinct intracellular molecules for signal transduction including "death domain" of FAS R and 55 kDa TNF- $\alpha$ R that participates in their cytotoxic effects. NGF/TNF R can bind both NGF and related factors as well as TNF ligands. Chemokine receptors are G protein-coupled, seven transmembrane (7TM, serpentine) domain receptors.

<sup>f</sup> The Duffy blood group antigen (DARC) is an erythrocyte receptor that can bind several different chemokines. It belongs to the immunoglobulin superfamily but characteristics of its signal transduction events remain unclear.

<p>VII. Growth factors</p> <p>1.1 SCF, M-CSF, PDGF-AA, AB, BB, FLT-3L, VEGF, SSV-PDGF</p> <p>1.2 FGF<math>\alpha</math>, FGF<math>\beta</math></p> <p>1.3 EGF, TGF-<math>\alpha</math>, VV-F19 (EGF-like)</p> <p>1.4 IGF-I, IGF-II, Insulin</p> <p>1.5 NGF, BDNF, NT-3, NT-4<sup>9</sup></p> <p>2. TGF-<math>\beta</math>1,<math>\beta</math>2,<math>\beta</math>3</p>	<p>VII. RKF</p> <p>1. TK sub-family</p> <p>1.1 IgTK III R</p> <p>1.2 IgTK IV R</p> <p>1.3 Cysteine-rich TK-I</p> <p>1.4 Cysteine rich TK-II</p> <p>1.5 Cysteine knot TK V</p> <p>2. STK subfamily<sup>h</sup></p>
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Exemplary peptides for this invention appear in Tables 4 through 20 below. These peptides may be prepared by methods disclosed in the art. Single letter amino acid abbreviations are used. The X in these sequences (and throughout this specification, unless specified otherwise in a particular instance) means that any of the 20 naturally occurring amino acid residues may be present. Any of these peptides may be linked in tandem (i.e., sequentially), with or without linkers, and a few tandem-linked examples are provided in the table. Linkers are listed as "A" and may be any of the linkers described herein. Tandem repeats and linkers are shown separated by dashes for clarity. Any peptide containing a cysteinyl residue may be cross-linked with another Cys-containing peptide, either or both of which may be linked to a vehicle. A few cross-linked examples are provided in the table. Any peptide having more than one Cys residue may form an intrapeptide disulfide bond, as well; see, for example, EPO-mimetic peptides in Table 5. A few examples of intrapeptide disulfide-bonded peptides are specified in the table. Any of these peptides may be derivatized as described herein, and a few derivatized examples are provided in the table. Derivatized peptides in

<sup>9</sup> The neurotrophic cytokines can associate with NGF/TNF receptors also.

the tables are exemplary rather than limiting, as the associated underivatized peptides may be employed in this invention, as well. For derivatives in which the carboxyl terminus may be capped with an amino group, the capping amino group is shown as  $-NH_2$ . For derivatives in which amino acid residues are substituted by moieties other than amino acid residues, the substitutions are denoted by  $\sigma$ , which signifies any of the moieties described in Bhatnagar *et al.* (1996), *J. Med. Chem.* 39: 3814-9 and Cuthbertson *et al.* (1997), *J. Med. Chem.* 40: 2876-82, which are incorporated by reference. The J substituent and the Z substituents ( $Z_5, Z_6, \dots Z_{40}$ ) are as defined in U.S. Pat. Nos. 5,608,035, 5,786,331, and 5,880,096, which are incorporated by reference. For the EPO-mimetic sequences (Table 5), the substituents  $X_2$  through  $X_{11}$  and the integer "n" are as defined in WO 96/40772, which is incorporated by reference. The substituents " $\Psi$ ," " $\Theta$ ," and "+" are as defined in Sparks *et al.* (1996), *Proc. Natl. Acad. Sci.* 93: 1540-4, which is hereby incorporated by reference.  $X_4, X_5, X_6$ , and  $X_7$  are as defined in U.S. Pat. No. 5,773,569, which is hereby incorporated by reference, except that: for integrin-binding peptides,  $X_1, X_2, X_3, X_4, X_5, X_6, X_7$ , and  $X_8$  are as defined in International applications WO 95/14714, published June 1, 1995 and WO 97/08203, published March 6, 1997, which are also incorporated by reference; and for VIP-mimetic peptides,  $X_1, X_1', X_1'', X_2, X_3, X_4, X_5, X_6$  and Z and the integers m and n are as defined in WO 97/40070, published October 30, 1997, which is also incorporated by reference. Xaa and Yaa below are as defined in WO 98/09985, published March 12, 1998, which is incorporated by reference.  $AA_1, AA_2, AB_1, AB_2$ , and AC are as defined in International application WO 98/53842, published December 3, 1998, which is incorporated by reference.  $X^1, X^2, X^3$ , and  $X^4$  in Table 17 only are as defined in European application EP 0 911

<sup>a</sup> STKS may encompass many other TGF- $\beta$ -related factors that remain unassigned. The protein kinases are intrinsic part of the intracellular domain of receptor kinase family (RKF). The enzymes participate in the signals transmission via the receptors.

393, published April 28, 1999. Residues appearing in boldface are D-amino acids. All peptides are linked through peptide bonds unless otherwise noted. Abbreviations are listed at the end of this specification. In the "SEQ ID NO." column, "NR" means that no sequence listing is required for the given sequence.

Table 4—IL-1 antagonist peptide sequences

Sequence/structure	SEQ ID NO:
<b>Z</b> <sub>11</sub> <b>Z</b> <sub>7</sub> <b>Z</b> <sub>8</sub> <b>QZ</b> <sub>5</sub> <b>YZ</b> <sub>6</sub> <b>Z</b> <sub>9</sub> <b>Z</b> <sub>10</sub>	212
<b>XXQZ</b> <sub>5</sub> <b>YZ</b> <sub>6</sub> <b>XX</b>	907
<b>Z</b> <sub>7</sub> <b>XQZ</b> <sub>5</sub> <b>YZ</b> <sub>6</sub> <b>XX</b>	908
<b>Z</b> <sub>7</sub> <b>Z</b> <sub>8</sub> <b>QZ</b> <sub>5</sub> <b>YZ</b> <sub>6</sub> <b>Z</b> <sub>9</sub> <b>Z</b> <sub>10</sub>	909
<b>Z</b> <sub>11</sub> <b>Z</b> <sub>7</sub> <b>Z</b> <sub>8</sub> <b>QZ</b> <sub>5</sub> <b>YZ</b> <sub>6</sub> <b>Z</b> <sub>9</sub> <b>Z</b> <sub>10</sub>	910
<b>Z</b> <sub>12</sub> <b>Z</b> <sub>13</sub> <b>Z</b> <sub>14</sub> <b>Z</b> <sub>15</sub> <b>Z</b> <sub>16</sub> <b>Z</b> <sub>17</sub> <b>Z</b> <sub>18</sub> <b>Z</b> <sub>19</sub> <b>Z</b> <sub>20</sub> <b>Z</b> <sub>21</sub> <b>Z</b> <sub>22</sub> <b>Z</b> <sub>11</sub> <b>Z</b> <sub>7</sub> <b>Z</b> <sub>8</sub> <b>QZ</b> <sub>5</sub> <b>YZ</b> <sub>6</sub> <b>Z</b> <sub>9</sub> <b>Z</b> <sub>10</sub> <b>L</b>	917
<b>Z</b> <sub>23</sub> <b>NZ</b> <sub>24</sub> <b>Z</b> <sub>25</sub> <b>Z</b> <sub>26</sub> <b>Z</b> <sub>27</sub> <b>Z</b> <sub>28</sub> <b>Z</b> <sub>29</sub> <b>Z</b> <sub>30</sub> <b>Z</b> <sub>40</sub>	979
TANVSSFEWTPYYWQPYALPL	213
SWTDYGYWQPYALPISGL	214
ETPFTWEESNAYYWQPYALPL	215
ENTYSPNWADSMYWQPYALPL	216
SVGEDHNFWTSEYWQPYALPL	217
DGYDRWRQSGERYWQPYALPL	218
FEWTPGYWQPY	219
FEWTPGYWQHY	220
FEWTPGWYQJY	221
AcFEWTPGWYQJY	222
FEWTPGWpYQJY	223
FAWTPGYWQJY	224
FEWAPGYWQJY	225
FEWVPGYWQJY	226
FEWTPGYWQJY	227
AcFEWTPGYWQJY	228
FEWTPaWYQJY	229
FEWTPSarWYQJY	230
FEWTPGYYPY	231
FEWTPGWWQPY	232
FEWTPNYWQPY	233
FEWTPvYWQJY	234
FEWTPecGYWQJY	235
FEWTPAibYWQJY	236
FEWTSarGYWQJY	237
FEWTPGYWQPY	238
FEWTPGYWQHY	239
FEWTPGWYQJY	240

AcFEWTPGWWYQJY	241
FEWTPGW-pY-QJY	242
FAWTPGYWQJY	243
FEWAPGYWQJY	244
FEWVPGYWQJY	245
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AcFEWTPGYWQJY	247
FEWTPAWYQJY	248
FEWTPSarWYQJY	249
FEWTPGYYQPY	250
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FEWTPVYWQJY	253
FEWTPecGYWQJY	254
FEWTPAibYWQJY	255
FEWTSarGYWQJY	256
FEWTPGYWQPYALPL	257
1NapEWTPGYYQJY	258
YEWTPGYYQJY	259
FEWVPGYYQJY	260
FEWTPSYYQJY	261
FEWTPNYYQJY	262
TKPR	263
RKSSK	264
RKQDK	265
NRKQDK	266
RKQDKR	267
ENRKQDKRF	268
VTKFYF	269
VTKFY	270
VTDFY	271
SHLYWQPYSVQ	671
TLVYWQPYSLQT	672
RGDYWQPYSVQS	673
VHVYWQPYSVQT	674
RLVYWQPYSVQT	675
SRVWFQPYSLQS	676
NMVDYWQPYSIQT	677
SVVFWQPYSVQT	678
TFVYWQPYALPL	679
TLVYWQPYSIQR	680
RLVYWQPYSVQR	681
SPVFWQPYSIQI	682
WIEWWQPYSVQS	683
SLIYWQPYSLQM	684
TRLYWQPYSVQR	685
RCDYWQPYSVQT	686
MRVFWQPYSVQN	687
KIVYWQPYSVQT	688
RHLYWQPYSVQR	689



ALVWWQPYSEI	690
SRVWFQPYSLQS	691
WEQPYALPLE	692
QLVWWQPYSVQR	693
DLRYWQPYSVQV	694
ELVWWQPYSLQL	695
DLVWWQPYSVQW	696
NGNYWQPYSFQV	697
ELVYWQPYSIQR	698
ELMYWQPYSVQE	699
NLLYWQPYSMQD	700
GYPEWQPYSVQR	701
SRVWYQPYSVQR	702
LSEQYQPYSVQR	703
GGGWWQPYSVQR	704
VGRWYQPYSVQR	705
VHVVWQPYSVQR	706
QARWYQPYSVQR	707
VHVVWQPYSVQT	708
RSVWYQPYSVQR	709
TRVWFQPYSVQR	710
GRIWFQPYSVQR	711
GRVWFQPYSVQR	712
ARTWYQPYSVQR	713
ARVWWQPYSVQM	714
RLMFYQPYSVQR	715
ESMWYQPYSVQR	716
HFGWWQPYSVHM	717
ARFWWQPYSVQR	718
RLVYWQ PYAPIY	719
RLVYWQ PYSYQT	720
RLVYWQ PYSLPI	721
RLVYWQ PYSVQA	722
SRVWYQ PYAKGL	723
SRVWYQ PYAQGL	724
SRVWYQ PYAMPL	725
SRVWYQ PYSVQA	726
SRVWYQ PYSLGL	727
SRVWYQ PYAREL	728
SRVWYQ PYSRQP	729
SRVWYQ PYFVQP	730
EYEWYQ PYALPL	731
IPEYWQ PYALPL	732
SRIWWQ PYALPL	733
DPLFWQ PYALPL	734
SRQWVQ PYALPL	735
IRSWWQ PYALPL	736
RGYWQ PYALPL	737
RLLWVQ PYALPL	738
EYRWFQ PYALPL	739

DAYWVQ PYALPL	740
WSGYFQ PYALPL	741
NIEFWQ PYALPL	742
TRDWVQ PYALPL	743
DSSWYQ PYALPL	744
IGNWYQ PYALPL	745
NLRWDQ PYALPL	746
LPEFWQ PYALPL	747
DSYWWQ PYALPL	748
RSQYYQ PYALPL	749
ARFWLQ PYALPL	750
NSYFWQ PYALPL	751
RFMYWQPYSVQR	752
AHLFWQPYSVQR	753
WWQPYALPL	754
YYQPYALPL	755
YFQPYALGL	756
YWYQPYALPL	757
RWWQPYATPL	758
GWYQPYALGF	759
YWYQPYALGL	760
IWYQPYAMPL	761
SNMQPYQRLS	762
TFVYWQPY AVGLPAAETACN	763
TFVYWQPY SVQMTITGKVTM	764
TFVYWQPY SSHXXVPXGFPL	765
TFVYWQPY YGNPQWAIHVRH	766
TFVYWQPY VLLELPEGAVRA	767
TFVYWQPY VDYVWPIPIAQV	768
GWYQPYVDGWR	769
RWEQPYVKDGWS	770
EWYQPYALGWAR	771
GWWQPYARGL	772
LFEQPYAKALGL	773
GWEQPYARGLAG	774
AWWQPYATPLDE	775
MWYQPYSSQPAE	776
GWTQPYSSQGEV	777
DWFQPYSIQSDE	778
PWQPYARGFG	779
RPLYWQPYSVQV	780
TLIYWQPYSVQI	781
RFDYWQPYSQDT	782
WHQFVQPYALPL	783
EWDS VYWQPYSVQ TLLR	784
WEQN VYWQPYSVQ SFAD	785
SDV VYWQPYSVQ SLEM	786
YYDG VYWQPYSVQ VMPA	787
SDIWYQ PYALPL	788
QRIWWQ PYALPL	789

SRIWWQ PYALPL	790
RSLYWQ PYALPL	791
TIIWEQ PYALPL	792
WETWYQ PYALPL	793
SYDWEQ PYALPL	794
SRIWCQ PYALPL	795
EIMFWQ PYALPL	796
DYVWQQ PYALPL	797
MDLLVQ WYQPYALPL	798
GSKVIL WYQPYALPL	799
RQGANI WYQPYALPL	800
GGGDEP WYQPYALPL	801
SQLERT WYQPYALPL	802
ETWVRE WYQPYALPL	803
KKGSTQ WYQPYALPL	804
LQARMN WYQPYALPL	805
EPRSQK WYQPYALPL	806
VKQKWR WYQPYALPL	807
LRRHDV WYQPYALPL	808
RSTASI WYQPYALPL	809
ESKEDQ WYQPYALPL	810
EGLTMK WYQPYALPL	811
EGSREG WYQPYALPL	812
VIEWWQ PYALPL	813
VWYWEQ PYALPL	814
ASEWWQ PYALPL	815
FYEWVQ PYALPL	816
EGWWVQ PYALPL	817
WGEWLQ PYALPL	818
DYVWEQ PYALPL	819
AHTWWQ PYALPL	820
FIEWFQ PYALPL	821
WLAWEQ PYALPL	822
VMEWWQ PYALPL	823
ERMWQ PYALPL	824
NXXWXX PYALPL	825
WGNWYQ PYALPL	826
TLYWEQ PYALPL	827
VWRWEQ PYALPL	828
LLWTQ PYALPL	829
SRIWXX PYALPL	830
SDIWYQ PYALPL	831
WGYYXX PYALPL	832
TSGWYQ PYALPL	833
VHPYXX PYALPL	834
EHSYFQ PYALPL	835
XXIWYQ PYALPL	836
AQLHSQ PYALPL	837
WANWFQ PYALPL	838
SRLYSQ PYALPL	839

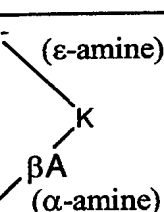
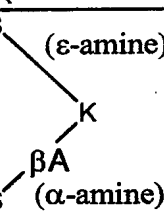
GVTFSQ PYALPL	840
SIVWSQ PYALPL	841
SRDLVQ PYALPL	842
HWGH VYWQPYSVQ DDLG	843
SWHS VYWQPYSVQ SVPE	844
WRDS VYWQPYSVQ PESA	845
TWDA VYWQPYSVQ KWLD	846
TPPW VYWQPYSVQ SLDP	847
YWSS VYWQPYSVQ SVHS	848
YWy QPY ALGL	849
YWy QPY ALPL	850
EWI QPY ATGL	851
NWE QPY AKPL	852
AFY QPY ALPL	853
FLY QPY ALPL	854
VCK QPY LEWC	855
ETPFTWEESNAYYWQPYALPL	856
QGWLTWQDSVDMYWQPYALPL	857
FSEAGYTPENTYWQPYALPL	858
TESPGGLDWAKIYWQPYALPL	859
DGYDRWRQSGERYYWQPYALPL	860
TANVSSFEWTPGYWQPYALPL	861
SVGEDHNFWTSE YWQPYALPL	862
MNDQTSEVSTFP YWQPYALPL	863
SWSEAFEQPRNL YWQPYALPL	864
QYAEPSALNDWG YWQPYALPL	865
NGDWATADWSNY YWQPYALPL	866
THDEHI YWQPYALPL	867
MLEKTYTTWTPG YWQPYALPL	868
WSDPLTRDADL YWQPYALPL	869
SDAFTTQDSQAM YWQPYALPL	870
GDDAAWRTDSTL YWQPYALPL	871
AIIRQLYRWSEM YWQPYALPL	872
ENTYSPNWADSM YWQPYALPL	873
MNDQTSEVSTFP YWQPYALPL	874
SVGEDHNFWTSE YWQPYALPL	875
QTPFTWEESNAY YWQPYALPL	876
ENPFTWQESNAY YWQPYALPL	877
VTPFTWEDSNVF YWQPYALPL	878
QIPFTWEQSNAY YWQPYALPL	879
QAPLTWQESAAY YWQPYALPL	880
EPTFTWEESKAT YWQPYALPL	881
TTTLTWEESNAY YWQPYALPL	882
ESPLTWEESAL YWQPYALPL	883
ETPLTWEESNAY YWQPYALPL	884
EATFTWAESNAY YWQPYALPL	885
EALFTWKESTAY YWQPYALPL	886
STP-TWEESNAY YWQPYALPL	887
ETPFTWEESNAY YWQPYALPL	888
KAPFTWEESQAY YWQPYALPL	889

STSFTWEESNAY YWQPYALPL	890
DSTFTWEESNAY YWQPYALPL	891
YIPFTWEESNAY YWQPYALPL	892
QTAFTWEESNAY YWQPYALPL	893
ETLFTWEESNAT YWQPYALPL	894
VSSFTWEESNAY YWQPYALPL	895
QPYALPL	896
Py-1-NapPYQJYALPL	897
TANVSSFEWTPG YWQPYALPL	898
FEWTPGYWQPYALPL	899
FEWTPGYWQJYALPL	900
FEWTPGYYQJYALPL	901
ETPFTWEESNAYYWQPYALPL	902
FTWEESNAYYWQJYALPL	903
ADVL YWQPYA PVT LWV	904
GDVAE YWQPYA LPLTSL	905
SWTDYG YWQPYA LPISGL	906
FEWTPGYWQPYALPL	911
FEWTPGYWQJYALPL	912
FEWTPGWYQPYALPL	913
FEWTPGWYQJYALPL	914
FEWTPGYYQPYALPL	915
FEWTPGYYQJYALPL	916
TANVSSFEWTPGYWQPYALPL	918
SWTDYGYWQPYALPISGL	919
ETPFTWEESNAYYWQPYALPL	920
ENTYSPNWADSMYWQPYALPL	921
SVGEDHNFWTSEYWQPYALPL	922
DGYDRWRQSGERYWQPYALPL	923
FEWTPGYWQPYALPL	924
FEWTPGYWQPY	925
FEWTPGYWQJY	926
EWTPGYWQPY	927
FEWTPGWYQJY	928
AEWTPGYWQJY	929
FAWTPGYWQJY	930
FEATPGYWQJY	931
FEWAPGYWQJY	932
FEWTAGYWQJY	933
FEWTPAYWQJY	934
FEWTPGAWQJY	935
FEWTPGYAQJY	936
FEWTPGYWQJA	937
FEWTGGYWQJY	938
FEWTPGYWQJY	939
FEWTJGYWQJY	940
FEWTPecGYWQJY	941
FEWTPAibYWQJY	942
FEWTPSarWYQJY	943
FEWTSarGYWQJY	944

FEWTPNYWQJY	945
FEWTPVYWQJY	946
FEWTPYVWQJY	947
AcFEWTPGWYQJY	948
AcFEWTPGYWQJY	949
INap-EWTPGYYQJY	950
YEWTPGYYQJY	951
FEWVPGYYQJY	952
FEWTPGYYQJY	953
FEWTPsYYQJY	954
FEWTPnYYQJY	955
SHLY-Nap-QPYSVQM	956
TLVY-Nap-QPYSLQT	957
RGDY-Nap-QPYSVQS	958
NMVY-Nap-QPYSIQT	959
VYWQPYSVQ	960
VY-Nap-QPYSVQ	961
TFVYWQJYALPL	962
FEWTPGYYQJ-Bpa	963
XaaFEWTPGYYQJ-Bpa	964
FEWTPGY-Bpa-QJY	965
AcFEWTPGY-Bpa-QJY	966
FEWTPG-Bpa-YQJY	967
AcFEWTPG-Bpa-YQJY	968
AcFE-Bpa-TPGYYQJY	969
AcFE-Bpa-TPGYYQJY	970
Bpa-EWTPGYYQJY	971
AcBpa-EWTPGYYQJY	972
VYWQPYSVQ	973
RLVYWQPYSVQR	974
RLVY-Nap-QPYSVQR	975
RLDYWQPYSVQR	976
RLVWFQPYSVQR	977
RLVYWQPYSIQR	978
DNSSWYDSFLL	980
DNTAWYESFLA	981
DNTAWYENFLL	982
PARE DNTAWYDSFLI WC	983
TSEY DNTTWYKFLA SQ	984
SQIP DNTAWYQSFL HG	985
SPFI DNTAWYENFLL TY	986
EQIY DNTAWYDHFL SY	987
TPFI DNTAWYENFLL TY	988
TYTY DNTAWYERFLM SY	989
TMTQ DNTAWYENFLL SY	990
TI DNTAWYANLVQ TYPQ	991
TI DNTAWYERFLA QYPD	992
HI DNTAWYENFLL TYTP	993
SQ DNTAWYENFLL SYKA	994
QI DNTAWYERFLL QYNA	995

NQ DNTAWYESFLL QYNT	996
TI DNTAWYENFLL NHNL	997
HY DNTAWYERFLQ QGWH	998
ETPFTWEESNAYYWQPYALPL	999
YIPFTWEESNAYYWQPYALPL	1000
DGYDRWRQSGERYWQPYALPL	1001
pY-INap-pY-QJYALPL	1002
TANVSSFEWTPGYWQPYALPL	1003
FEWTPGYWQJYALPL	1004
FEWTPGYWQPYALPLSD	1005
FEWTPGYYQJYALPL	1006
FEWTPGYWQJY	1007
AcFEWTPGYWQJY	1008
AcFEWTPGWYQJY	1009
AcFEWTPGYYQJY	1010
AcFEWTPaYWQJY	1011
AcFEWTPaWYQJY	1012
AcFEWTPaYYQJY	1013
FEWTPGYYQJYALPL	1014
FEWTPGYWQJYALPL	1015
FEWTPGWYQJYALPL	1016
TANVSSFEWTPGYWQPYALPL	1017
AcFEWTPGYWQJY	1018
AcFEWTPGWYQJY	1019
AcFEWTPGYYQJY	1020
AcFEWTPAYWQJY	1021
AcFEWTPAWYQJY	1022
AcFEWTPAYYQJY	1023

Table 5—EPO-mimetic peptide sequences

Sequence/structure	SEQ ID NO:
YXCXXGPXTWXCXP	83
YXCXXGPXTWXCXP-YXCXXGPXTWXCXP	84
YXCXXGPXTWXCXP- $\Lambda$ -YXCXXGPXTWXCXP	85
YXCXXGPXTWXCXP- $\Lambda$ -  ( $\epsilon$ -amine) K $\beta$ A ( $\alpha$ -amine)	86
GGTYSCHFGPLTWVCKPQGG	87
GGDYHCRMGPLTWVCKPLGG	88
GGVYACRMGPITWVCSPLGG	89
VGNYMCHFGPITWVCRPGGG	90
GGLYLCRFGPVTWDCGYKGG	91
GGTYSCHFGPLTWVCKPQGG-	92
GGTYSCHFGPLTWVCKPQGG	92
GGTYSCHFGPLTWVCKPQGG- $\Lambda$ -	93
GGTYSCHFGPLTWVCKPQGG	93
GGTYSCHFGPLTWVCKPQGGSSK	94
GGTYSCHFGPLTWVCKPQGGSSK-	95
GGTYSCHFGPLTWVCKPQGGSSK	95
GGTYSCHFGPLTWVCKPQGGSSK- $\Lambda$ -	96
GGTYSCHFGPLTWVCKPQGGSSK	96
GGTYSCHFGPLTWVCKPQGGSS  ( $\epsilon$ -amine) K $\beta$ A ( $\alpha$ -amine)	97
GGTYSCHFGPLTWVCKPQGGSS	97
GGTYSCHFGPLTWVCKPQGGSSK(- $\Lambda$ -biotin)	98
CX <sub>4</sub> X <sub>5</sub> GPX <sub>6</sub> TWX <sub>7</sub> C	421
GGTYSCHGPLTWVCKPQGG	422
VGNVMAHMGPIWVCRPGG	423
GGPHHVYACRMGPLTWIC	424
GGTYSCHFGPLTWVCKPQ	425
GGLYACHMGPMTWVCQPLRG	426
TIAQYICYMGPETWECRSPKA	427
YSCHFGPLTWVCK	428
YCHFGPLTWVC	429
X <sub>2</sub> X <sub>3</sub> X <sub>4</sub> GPX <sub>5</sub> TWX <sub>6</sub> X <sub>7</sub>	124
YX <sub>2</sub> X <sub>3</sub> X <sub>4</sub> GPX <sub>5</sub> TWX <sub>6</sub> X <sub>7</sub>	461



$X_1 Y X_2 X_3 X_4 X_5 G P X_6 T W X_7 X_8 X_9 X_{10} X_{11}$	419
$X_1 Y X_2 C X_3 X_4 G P X_5 T W X_6 C X_7 X_8 X_9 X_{10} X_{11}$	420
GGLYLCRFGPVTWDCGYKGG	1024
GGTYSCHFGPLTWVCKPQGG	1025
GGDYHCRMGPLTWVCKPLGG	1026
VGNYMCHFGPITWVCRPGGG	1029
GGVYACRMGPITWVCSPLGG	1030
VGNYMAHMGPIWVCRPGG	1035
GGTYSCHFGPLTWVCKPQ	1036
GGLYACHMGPMTWVCQPLRG	1037
TIAQYICYMGPETWECPSPKA	1038
YSCHFGPLTWVCK	1039
YCHFGPLTWVC	1040
SCHFGPLTWVCK	1041
$(A X_2) X_3 X_4 X_5 G P X_6 T W X_7 X_8$	1042

Table 6—TPO-mimetic peptide sequences

Sequence/structure	SEQ ID NO:
IEGPTLRQWLAARA	13
IEGPTLRQWLAACA	24
IEGPTLRQWLAARA	25
IEGPTLRQWLAARA- $\Delta$ -IEGPTLRQWLAARA	26
IEGPTLRQWLAACA- $\Delta$ -IEGPTLRQWLAACA	27
IEGPTLRQCLAARA- $\Delta$ -IEGPTLRQCLAARA	28
IEGPTLRQWLAARA- $\Delta$ -K(BrAc)- $\Delta$ -IEGPTLRQWLAARA	29
IEGPTLRQWLAARA- $\Delta$ -K(PEG)- $\Delta$ -IEGPTLRQWLAARA	30
IEGPTLRQCLAARA- $\Delta$ -IEGPTLRQWLAARA	31
IEGPTLRQCLAARA- $\Delta$ -IEGPTLRQWLAARA	31
IEGPTLRQWLAARA- $\Delta$ -IEGPTLRQCLAARA	32
IEGPTLRQWLAARA- $\Delta$ -IEGPTLRQCLAARA	32
VRDQIXXXL	33
TLREWL	34
GRVRDQVAGW	35
GRVKDQIAQL	36
GVRDQVSWAL	37
ESVREQVMKY	38
SVRSQISASL	39
GVRETVYRHM	40
GVREVIVMHML	41
GRVRDQIWAAL	42
AGVRDQILIWL	43
GRVRDQIMLSL	44
GRVRDQI(X) <sub>2</sub> L	45
CTLRQWLQGC	46
CTLQEFLEG	47
CTRTEWLHGC	48
CTLREWLHGGFC	49
CTLREWVFAGLC	50
CTLRQWLILLGMC	51
CTLAEFASGVEQC	52
CSLQEFSLHGGYVC	53
CTLREFLDPTTAVC	54
CTLKEWLVSHEVWC	55
CTLREWL(X) <sub>2,6</sub> C	56-60
REGPTLRQWM	61
EGPTLRQWLA	62
ERGPFWAKAC	63
REGPRCVMWM	64
CGTEGPTLSTWLDC	65

CEQDGPTLLEWLKC	66
CELVGPSLMSWLTC	67
CLTGPFVTQWLYEC	68
CRAGPTLLEWLTLC	69
CADGPTLREWISFC	70
C(X) <sub>1,2</sub> EGPTLREWL(X) <sub>1,2</sub> C	71-74
GGCTLREWLHGGFCGG	75
GGCADGPTLREWISFCGG	76
GNADGPTLRQWLEGRRPKN	77
LAIEGPTLRQWLHGNGRDT	78
HGRVGPTLREWKTQVATKK	79
TIKGPTLRQWLKSREHTS	80
ISDGPTLKEWLSVTRGAS	81
SIEGPTLREWLTSTRTPHS	82

Table 7—G-CSF-mimetic peptide sequences

Sequence/structure	SEQ ID NO:
EEDCK	99
EEDCK	99
 EEDCK	99
EED $\sigma$ K	100
EED $\sigma$ K	100
 EED $\sigma$ K	100
pGluED $\sigma$ K	101
pGluED $\sigma$ K	101
 pGluED $\sigma$ K	101
PicSD $\sigma$ K	102
PicSD $\sigma$ K	102
 PicSD $\sigma$ K	102
EEDCK- $\Lambda$ -EEDCK	103
EEDXK- $\Lambda$ -EEDXK	104

Table 8—TNF-antagonist peptide sequences

Sequence/structure	SEQ ID NO:
YCFTASENHCY	106
YCFTNSENHCY	107
YCFTRSENHCY	108
FCASENHCY	109
YCASENHCY	110
FCNSENHCY	111
FCNSENRCY	112
FCNSVENRCY	113
YCSQSVSND CF	114
FCVSNDRCY	115
YCRKELGQVCY	116
YCKEPGQCY	117
YCRKEMGCY	118
FCRKEMGCY	119
YCWSQNLCY	120
YCELSQYLCY	121
YCWSQNYCY	122
YCWSQYLCY	123
DFLPHYKNTSLGHRP	1085
$  \begin{array}{c}  AA_1-AB_1 \\  \quad \quad \backslash \\  \quad \quad \quad AC \\  \quad \quad / \\  AA_2-AB_2  \end{array}  $	NR

Table 9—Integrin-binding peptide sequences

Sequence/structure	SEQ ID NO:
RX,ETX,WX <sub>3</sub>	441
RX,ETX,WX <sub>3</sub>	442
RGDGX	443
CRGDGXC	444
CX,X <sub>2</sub> RLDX <sub>3</sub> X <sub>4</sub> C	445
CARRLDAPC	446
CPSRLDSPC	447
X,X <sub>2</sub> X <sub>3</sub> RGDX <sub>4</sub> X <sub>5</sub> X <sub>6</sub>	448
CX <sub>2</sub> CRGDCX <sub>2</sub> C	449
CDCRGDCFC	450
CDCRGDCLC	451
CLCRGDCIC	452
X,X <sub>2</sub> DDX <sub>4</sub> X <sub>5</sub> X <sub>6</sub>	453
X,X <sub>2</sub> X <sub>3</sub> DDX <sub>4</sub> X <sub>5</sub> X <sub>6</sub> X <sub>7</sub> X <sub>8</sub>	454
CWDDGWLC	455
CWDDLWWLC	456
CWDDGLMC	457
CWDDGWMC	458
CSWDDGWLC	459
CPDDLWWLC	460
NGR	NR
GSL	NR
RGD	NR
CGRECPRLCQSSC	1071
CNGRCVSGCAGRC	1072
CLSGSLSC	1073
RGD	NR
NGR	NR
GSL	NR
NGRAHA	1074
CNGRC	1075
CDCRGDCFC	1076
CGSLVRC	1077
DLXXL	1043
RTDLDSLRTYTL	1044
RTDLDSLRTY	1053
RTDLDSLRT	1054
RTDLDSLRL	1078
GDLDLLKLRLTL	1079
GDLHSLRQLLSR	1080
RDDLHMLRLQLW	1081
SSDLHALKKRYG	1082
RGDLKQLSELTW	1083
RGDLAALSAPPV	1084

Table 10—Selectin antagonist peptide sequences

Sequence/structure	SEQ ID NO:
DITWDQLWDLMK	147
DITWDELWKIMN	148
DYTWFEWDMMQ	149
QITWAQLWNMMK	150
DMTWHDLWTLMS	151
DYSWHDLWEMMS	152
EITWDQLWEVMN	153
HVSWEQLWDIMN	154
HITWDQLWRIMT	155
RNMSWLELWEHMK	156
AEWTWDQLWHVMNPAESQ	157
HRAEWLALWEQMSP	158
KKEDWLALWRIMSV	159
ITWDQLWDLMK	160
DITWDQLWDLMK	161
DITWDQLWDLMK	162
DITWDQLWDLMK	163
CQNRYTDLVAIQNKNE	462
AENWADNEPNNKRNNED	463
RKNNKTWTWVGTKKALTNE	464
KKALTNEAENWAD	465
CQXRYTDLVAIQNKXE	466
RKXNXXWTWVGTXKXLTEE	467
AENWADGEPNNKXNXED	468
CXXXYTXLVAIQNKXE	469
RKXXXXWXWVGTXKXLTXE	470
AXNWXXXEPNNXXXED	471
XKXKTXEAXNWXX	472

Table 11—Antipathogenic peptide sequences

Sequence/structure	SEQ ID NO:
GFFALIPKIISSPLFKTLLSAVGSALSSSSGGQQ	503
GFFALIPKIISSPLFKTLLSAVGSALSSSSGGQE	504
GFFALIPKIISSPLFKTLLSAV	505
GFFALIPKIISSPLFKTLLSAV	506
KGFFALIPKIISSPLFKTLLSAV	507
KKGFFALIPKIISSPLFKTLLSAV	508
KKGFFALIPKIISSPLFKTLLSAV	509
GFFALIPKIIIS	510
GIGAVLKVLTTGLPALISWIKRKRQQ	511
GIGAVLKVLTTGLPALISWIKRKRQQ	512
GIGAVLKVLTTGLPALISWIKRKRQQ	513
GIGAVLKVLTTGLPALISWIKR	514
AVLKVLTTGLPALISWIKR	515
KLLLLLKLLLLK	516
KLLLKLLLKLLK	517
KLLLKLLLKLLK	518
KKLLKLLKLLK	519
KLLLKLLLKLLK	520
KLLLKLLLKLLK	521
KLLLLK	522
KLLLKLLK	523
KLLLKLLKLLK	524
KLLLKLLKLLK	525
KLLLKLLKLLK	526
KAAAKAAAKAAK	527
KVVVKVVVKVVK	528
KVVVKVVKVVVK	529
KVVVKVVKVVVK	530
KVVVKVVKVVVK	531
KLILKL	532
KVLHLL	533
LKLRL	534
KPLHLL	535
KLILKLVR	536
KVFHLLHL	537
HKFRILKL	538
KPFHILHL	539
KIIKIKIKIK	540
KIIKIKIKIK	541
KIIKIKIKIK	542
KIPIKIKIPK	543
KIPIKIKIVK	544
RIIRIRIRIR	545
RIIRIRIRIR	546
RIIRIRIRIR	547
RIVIRIRIR	548



RIIVRIRLRIIR	549
RIGIRLVRRIIR	550
KIVIRIRIRLIR	551
RIAVKWRLRFIK	552
KIGWKLRVRIIR	553
KKIGWLIIRVRR	554
RIVIRIRIRLIRIR	555
RIIVRIRLRIIRVR	556
RIGIRLVRRIIRRV	557
KIVIRIRARLIRIRIR	558
RIIVKIRLRIIKKIRL	559
KIGIKARVRIIRVKII	560
RIIVHIRLRIIHHIRL	561
HIGIKAHVRIIRVHII	562
RIYVKIHLRYIKKIRL	563
KIGHKARVHIIRYKII	564
RIYVKPHPRYIKKIRL	565
KPGHKARPHIIRYKII	566
KIVIRIRIRLIRIRIRKIV	567
RIIVKIRLRIIKKIRLIKK	568
KIGWKLRVRIIRVKIGRLR	569
KIVIRIRIRLIRIRIRKIVKVKRIR	570
RFAVKIRLRIIKKIRLIKKIRKRVIK	571
KAGWKLRVRIIRVKIGRLRKIGWKKRVRIK	572
RIYVKPHPRYIKKIRL	573
KPGHKARPHIIRYKII	574
KIVIRIRIRLIRIRIRKIV	575
RIIVKIRLRIIKKIRLIKK	576
RIYVSKISYIKKIRL	577
KIVIFTRIRLTSIRIRSIV	578
KPIHKARPTIIRYKMI	579
cyclicCKGFFALIPKIISSPLFKTLLSAVC	580
CKKGFFALIPKIISSPLFKTLLSAVC	581
CKKKGFFALIPKIISSPLFKTLLSAVC	582
CyclicCRIVIRIRIRLIRIRC	583
CyclicCKPGHKARPHIIRYKIIIC	584
CyclicCRFAVKIRLRIIKKIRLIKKIRKRVIKC	585
KLLLKLLL KLLKC	586
KLLLKLLLKLLK	587
KLLLKLKLKLLKC	588
KLLLKLLLKLLK	589

Table 12—VIP-mimetic peptide sequences

Sequence/structure	SEQ ID NO:
HSDAVFYDNYTR LRKQMAVKKYLN SILN	590
Nle HSDAVFYDNYTR LRKQMAVKKYLN SILN	591
X <sub>1</sub> X <sub>1</sub> ' X <sub>1</sub> " X <sub>2</sub>	592
X <sub>3</sub> S X <sub>4</sub> LN	593
NH CH CO KKYX5 NH CH CO X6 <div style="display: flex; justify-content: space-around; width: 100%;"> <div style="text-align: center;"> <math>\begin{array}{c}   \\ (\text{CH}_2)_m \end{array}</math> </div> <div style="text-align: center;">Z</div> <div style="text-align: center;"> <math>\begin{array}{c}   \\ (\text{CH}_2)_n \end{array}</math> </div> </div>	594
KKYL	595
NSILN	596
KKYL	597
KKYA	598
AVKKYL	599
NSILN	600
KKYV	601
SILauN	602
KKYLNle	603
NSYLN	604
NSIYN	605
KKYLPPNSILN	606
LauKKYL	607
CapKKYL	608
KYL	NR
KKYNle	609
VKKYL	610
LNSILN	611
YLNSILN	612
KKYLN	613
KKYLNS	614
KKYLNSI	615
KKYLNSIL	616
KKYL	617
KKYDA	618
AVKKYL	619
NSILN	620
KKYV	621
SILauN	622
NSYLN	623
NSIYN	624
KKYLNle	625
KKYLPPNSILN	626
KKYL	627
KKYDA	628
AVKKYL	629
NSILN	630
KKYV	631
SILauN	632

LauKKYL	633
CapKKYL	634
KYL	NR
KYL	NR
KKYNie	635
VKKYL	636
LNSILN	637
YLNSILN	638
KKYLNie	639
KKYLN	640
KKYLNS	641
KKYLNSI	642
KKYLNSIL	643
KKKYLD	644
cyclicCKKYLC	645
CKKYLK     S-CH <sub>2</sub> -CO	646
KKYA	647
WWTDTGLW	648
WWTDDGLW	649
WWDTRGLWVWTI	650
FWGNDGIWLESG	651
DWDQFGLWRGAA	652
RWDDNGLWVVVL	653
SGMWSHYGIWMG	654
GGRWDQAGLWVA	655
KLWSEQGIWMGE	656
CWSMHGLWLC	657
GCWDNTGIWVPC	658
DWDTRGLWVY	659
SLWDENGAWI	660
KWDDRGLWMH	661
QAWNERGLWT	662
QWDTRGLWVA	663
WNVHGIWQE	664
SWDTRGLWVE	665
DWDTRGLWVA	666
SWGRDGLWIE	667
EWDNGLWAL	668
SWDEKGLWSA	669
SWDSSGLWMD	670

Table 13—Mdm/hdm antagonist peptide sequences

Sequence/structure	SEQ ID NO:
TFSDLW	130
QETFSDLWKLLP	131
QPTFSDLWKLLP	132
QETFSDYWKLLP	133
QPTFSDYWKLLP	134
MPRFMDYWEGLN	135
VQNFIDYWTQQF	136
TGPAFTHYWATF	137
IDRAPTFRDHWFALV	138
PRPALVFADYWETLY	139
PAFSRFWSDLSAGAH	140
PAFSRFWSKLSAGAH	141
PXFXDYWXXL	142
QETFSDLWKLLP	143
QPTFSDLWKLLP	144
QETFSDYWKLLP	145
QPTFSDYWKLLP	146

Table 14—Calmodulin antagonist peptide sequences

Sequence/structure	SEQ ID NO:
SCVKWKGKEFCGS	164
SCWKYWGKECGS	165
SCYEWGKLRWCGS	166
SCLRWGKWSNCGS	167
SCWRWGKYQICGS	168
SCVSWGALKLCGS	169
SCIRWGQNTFCGS	170
SCWQWGNLKICGS	171
SCVRWGQLSICGS	172
LKKFNARRKLKGAILTTLAK	173
RRWKKNFIAVSAANRFKK	174
RKWQKTGHAVRAIGRLSS	175
INLKALAALAKKIL	176
KIWSILAPLGTTLVKLVA	177
LKKLLKLLKLLKL	178
LKWKKLLKLLKLLKLL	179
AEWPSLTEIKTLSHFSV	180
AEWPSPTRVISTTYFGS	181
AELAHWPPVKTVLRSFT	182
AEGSWLQLLNLMKQMN	183
AEWPSLTEIK	184